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Diagnostic Performance of Liquid-Based Versus Conventional Cytology In Body Fluid Examination: A Comparative Study

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ABSTRACT

Cytological examination of serous effusions is a valuable diagnostic tool for detecting malignancies, assessing tumour stage, and identifying inflammatory lesions. However, conventional smear techniques may be limited by low cellularity and obscuring background elements. Liquid-based cytology has emerged as a technique that may overcome these limitations. This study compares the cytomorphological features of body fluids processed by conventional smear and Liquid-based cytology methods.

AIM

The study compares the cytomorphological features of body fluids processed by conventional cytology and liquid-based cytology. It focuses on evaluating parameters such as cellularity, cell morphology, distribution, background, and architectural preservation for diagnostic accuracy.

MATERIALS AND METHODS

A cross-sectional analytical study was conducted on 124 body fluid samples, including pleural (n=53), peritoneal (n=53), and bronchial washings (n=18), collected between September 2023 and January 2025. Smears were prepared using both conventional cytology and Liquid-based cytology techniques and stained with Haematoxylin & Eosin, and Papanicolaou stains. Samples were evaluated for cellularity, cell morphology, distribution, background, and architecture using a standardized scoring system.

RESULTS

Liquid-based cytology smears consistently demonstrated superior performance in terms of cellularity, cell morphology, uniform distribution, and clear background when compared to conventional smears. These differences were statistically significant ($p < 0.05$) across all sample types, enhancing overall diagnostic yield and interpretability.

CONCLUSION

Liquid-based cytology offers clear advantages over conventional smears in the evaluation of body fluids, improving the quality and efficiency of cytological assessment. Its adoption in routine practice may aid in more accurate and rapid diagnoses, particularly in effusion cytology

Key Words: *Body fluids, Liquid-based cytology, Conventional cytology, Cytomorphology*

1. INTRODUCTION

Exfoliative cytology is a simple, minimally invasive diagnostic method that collects naturally shed epithelial cells from organ surfaces into body cavities. The pleural and peritoneal cavities are each lined by a single layer of flattened mesothelial cells, collectively referred to as the serosa. These cavities are anatomically divided into parietal and visceral layers, which are

separated by a potential space that normally contains only a minimal volume of lubricating fluid—pleural or peritoneal—ensuring smooth movement between surfaces. Under pathological conditions, this space may accumulate excessive fluid, resulting in what is termed as effusion [1].

Cytological examination of effusion fluids plays a pivotal role in the detection of malignancy, serving both as a valuable diagnostic modality and an indicator of disease dissemination. The identification of malignant cells in these fluids can significantly affect prognosis and disease staging. Due to its minimally invasive nature, cytology is often preferred over biopsy, offering a simpler, safer, and more cost-effective diagnostic approach [2,3–6].

Liquid-based cytology, initially developed for cervical screening and approved by the U.S. Food and Drug Administration in 1996, has since been adopted for cytological evaluation across various body sites, including serous effusions. This technique has shown promising diagnostic performance and is increasingly recognized as an alternative to conventional cytopreparatory methods [7,8,9].

In recent years, Liquid-based cytology has gained prominence owing to its ability to distribute cells in a monolayer on slides, which facilitates improved morphological evaluation. The method enhances cellular preservation and nuclear detail due to superior fixation, resulting in increased diagnostic accuracy compared to conventional smear techniques [1,10].

2. MATERIALS AND METHODS

This prospective cross-sectional study was conducted at SRM Medical College and Research Institute, Kattankulathur, from September 2023 to January 2025. A total of 124 body fluid samples—pleural fluid, peritoneal fluid, and bronchial wash—were selected based on inclusion criteria. Samples exceeding 50 ml in volume and received within 2 hours of collection were included. Samples were excluded if they were inadequate (<50 ml), unrefrigerated with delayed transport, or acellular smears.

Informed consent was obtained from all participants, and relevant clinical data, including age, sex, type of fluid, and clinical diagnosis, were recorded. Fluids were collected using standard procedures: pleural fluid via thoracocentesis, peritoneal fluid via abdominal tapping, and bronchial wash through bronchoscopy.

All specimens were collected in sterile containers and divided into two parts—one for conventional cytology and the other for liquid-based cytology. Gross characteristics such as volume, colour, and turbidity were documented. In the conventional method, samples were centrifuged at 1500–2000 rpm for 10–15 minutes. Smears were prepared from the sediment, stained with Haematoxylin and Eosin (H&E) and Papanicolaou (PAP) stains.

For Liquid-based cytology, the sediment was treated with lyser solution, fixed using Ezifix Cell Fixative, and processed using the EZIPREP NANOCYT NEO system, which created a monolayer smear. These smears were also stained with H&E and PAP stains.

Smears from both methods were evaluated based on five parameters: cellularity, cell morphology, cell distribution, background, and architectural preservation using the Mair et al. scoring system (Table 1). Diagnostic categorization followed The International System for Reporting Serous Effusion Cytopathology (TIS), classifying findings into nondiagnostic, negative for malignancy, atypia of undetermined significance (AUS), suspicious for malignancy, and malignant.

Table 1: The Mair et al. Scoring System - Parameters and Scores to evaluate conventional smears and Liquid-based cytology smears

PARAMETER	QUANTITATIVE AND QUALITATIVE DESCRIPTION	SCORE
Cellularity	Minimal, Diagnosis not possible Sufficient for diagnosis Abundant	0 1 2
Cell morphology	Poor Fair Good	0 1 2
Cell distribution	Random Uneven	0 1

	Even	2
Background	Great compromise in diagnosis Diagnosis possible Diagnosis easy	0 1 2
Architecture	Not preserved Partially preserved Well preserved	0 1 2

3. RESULTS

This analytical cross-sectional study included a total of 124 body fluid samples, comprising 53 pleural fluids, 53 peritoneal fluids, and 18 bronchial wash specimens. Both pleural and peritoneal fluids accounted for 42.74% of the cases each, while bronchial washes constituted 14.52%. Age-wise, most patients with pleural and peritoneal effusions were between 30 to 60 years, whereas bronchial wash samples were more commonly obtained from individuals above 60 years of age. In terms of gender distribution, peritoneal fluids showed a female predominance (58.5%), pleural fluids were almost evenly distributed between males and females, and bronchial washes were more common in males (66.7%).

All 124 samples were processed using both Conventional Smears and Liquid-Based Cytology techniques. Cytomorphological parameters assessed included cellularity, cell morphology, cell distribution, background, and architectural preservation. A scoring system adapted from Mair et al. was used for evaluation. Statistical analysis was performed using the **Wilcoxon signed-rank test**, which is suitable for comparing paired, non-parametric data. The results were interpreted based on positive ranks (favouring Liquid-based cytology), negative ranks (favouring Conventional smears), and ties (indicating equivalence). A p-value of <0.05 was considered statistically significant (Table 2).

Table 2: Statistical Analysis of All Morphological Parameters of Peritoneal, Pleural, and Bronchial Wash

Parameters	**Type of Fluid**	**Liquid-based cytology Superior**	**Conventional Superior**	**Equivalent**	**P-value**
Cellularity	Peritoneal	30 (56.6%)	4 (7.5%)	19 (35.9%)	0.041*
	Pleural	29 (54.7%)	6 (11.3%)	18 (34%)	0.004*
	Bronchial wash	8 (44.5%)	1 (5.5%)	9 (50%)	0.009*
Cell Morphology	Peritoneal	30 (56.6%)	5 (9.4%)	18 (34%)	0.032*
	Pleural	31 (58.5%)	4 (7.5%)	18 (34%)	0.002*
	Bronchial wash	9 (50%)	2 (11.1%)	7 (38.9%)	0.017*
Cell Distribution	Peritoneal	46 (86.8%)	2 (3.8%)	5 (9.4%)	0.008*
	Pleural	45 (84.9%)	3 (5.6%)	5 (9.4%)	< 0.001*
	Bronchial wash	12 (66.6%)	3 (16.7%)	3 (16.7%)	0.006*
Background	Peritoneal	31 (58.5%)	0 (0.0%)	22 (41.5%)	0.011*
	Pleural	30 (56.6%)	1 (1.9%)	22 (41.5%)	0.023*
	Bronchial wash	10 (55.6%)	1 (5.5%)	7 (38.9%)	0.003*

Architecture	Peritoneal	20 (37.7%)	5 (9.4%)	28 (52.9%)	0.015*
	Pleural	14 (26.4%)	4 (7.6%)	35 (66%)	0.008*
	Bronchial wash	4 (22.2%)	3 (16.7%)	11 (61.1%)	0.353

In terms of cellularity, Liquid-based cytology demonstrated superiority in 67 cases (54%) as compared to Conventional Smears, which was superior in 11 cases (8.9%), while 46 cases (37.1%) were considered equivalent (Figure 1). The difference in cellularity between the two techniques was statistically significant across all fluid types ($p < 0.05$). For cell morphology, Liquid-based cytology showed better morphology in 70 cases (56.5%) compared to Conventional smears, which showed better morphology in 11 cases (8.9%), with equivalent morphology noted in 43 cases (34.6%) (Figure 2). This difference also reached statistical significance ($p < 0.05$).

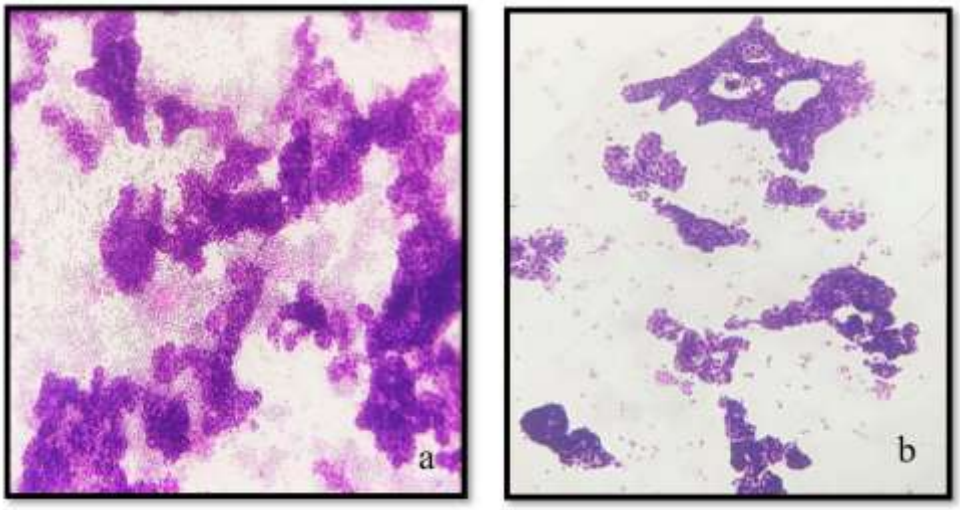

Figure 1. Cellularity: a.) Liquid-based cytology smear showing good cellularity in peritoneal fluid (H&E, 100X); b.) Conventional smear showing moderate cellularity in peritoneal fluid (H&E, 100X)

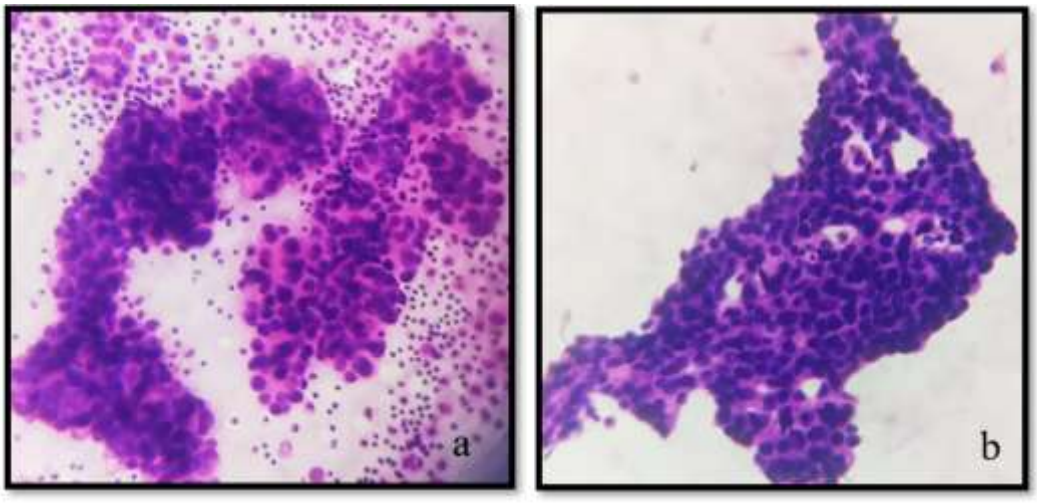

Figure 2. Cell morphology: a.) Liquid-based cytology smear cell morphology: Mesothelial cells arranged in clusters, Individual cells are round-oval, with a high N: C ratio, abundant cytoplasm, eccentrically pushed nucleus, with inconspicuous nucleoli- Suspicious of malignancy (H&E, 400X); b.) Conventional smear cell morphology: Mesothelial cells arranged in clusters. Individual cells are round-oval, with a high N: C ratio, moderate cytoplasm, eccentrically pushed nucleus-Suspicious of malignancy. (H&E, 400X)

Concerning cell distribution, Liquid-based cytology yielded a more uniform distribution in 103 cases (83%) compared to conventional smears, which were superior in 8 cases (6.5%), and 13 cases (10.5%) were equivalent. The difference was statistically significant across pleural, peritoneal, and bronchial samples. Background clarity was markedly better in Liquid-based cytology for 71 cases (57.3%), compared to only 2 cases (1.6%) for Conventional smear; 51 cases (41.1%) were equivalent (Figure 3). The difference in cellularity between the two techniques was statistically significant ($p < 0.05$).

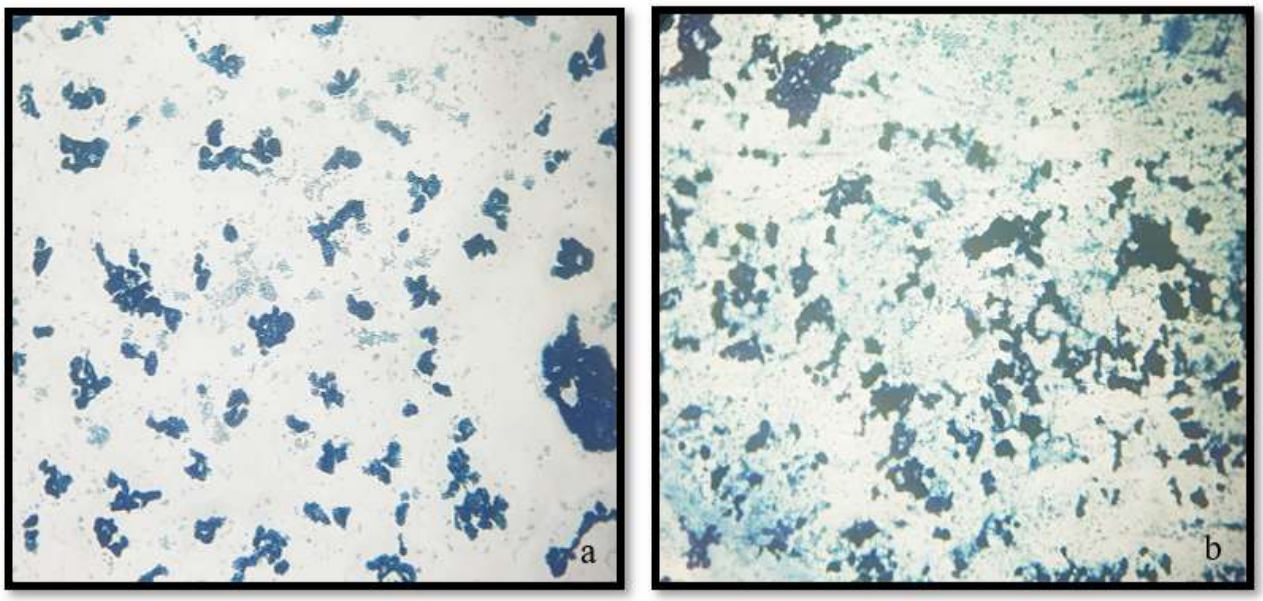

Figure 3. Background: a.) Liquid-based cytology smears showing clear background (PAP,40X); b.) Conventional smears showing proteinaceous background (PAP,40X)

Preservation of architecture, including nuclear-cytoplasmic ratio and three-dimensional clustering, was better appreciated in Liquid-based cytology for 38 cases (30.6%) compared to 12 cases (9.7%) for Conventional smear, with 74 cases (59.7%) showing equivalent features. Statistically significant differences were observed in pleural and peritoneal fluids ($p < 0.05$), whereas bronchial wash samples showed no significant difference ($p> 0.05$).

Diagnostic categorization based on The International System for Reporting Serous Effusion Cytopathology (TIS) indicated that among pleural fluids (n=53), 48 (90.6%) were negative, 1 (1.9%) suspicious, and 4 (7.5%) positive for malignancy. In peritoneal fluids (n=53), 39 (73.6%) were negative, 6 (11.3%) suspicious, and 8 (15.1%) positive. Bronchial washings (n=18),16 (88.9%) negative and 2 (11.1%) suspicious cases, with no positive findings. Peritoneal fluid had the highest rate of malignancy among the three types of fluid.

4. DISCUSSION

The cytological evaluation of serous fluids remains a cornerstone in diagnosing various benign and malignant conditions. In this comparative study, Liquid-Based Cytology demonstrated clear advantages over Conventional Smears across multiple parameters, with results largely concordant with findings reported in prior literature.

Liquid-Based Cytology yielded significantly higher cellularity in pleural, peritoneal, and bronchial wash fluids. This concordance with studies by Swathy et al. and Madhu et al. underscores Liquid-Based Cytology's ability to enhance cell yield, likely due to reduced cell loss and better sample preservation【11,12】.

Cell morphology was also better preserved in Liquid-Based Cytology, with enhanced nuclear and cytoplasmic detail facilitating improved diagnostic interpretation. This observation is concordant with studies by Swathy et al. and Siddiqui et al.【11,14】.

Uniform cell distribution—a hallmark of Liquid-Based Cytology —was consistently superior, reducing diagnostic pitfalls due to overlapping or clumped cells. These findings are in agreement with those of Alwahaibi et al. and Siddiqui et al., reflecting concordant evidence on Liquid-Based Cytology's technical efficiency in smearing and presentation【13,14】.

Background clarity was markedly improved in Liquid-Based Cytology preparations, which minimized obscuring elements like blood and proteinaceous debris. This again aligns concordantly with prior work, facilitating faster and more confident diagnoses【13,14】.

Concerning architectural preservation, Liquid-Based Cytology showed superiority in pleural and peritoneal fluids, allowing better visualization of cellular clusters and spatial arrangements. This is concordant with observations by Nemade et al., Amiri et al., and Ruchita et al.【15,16,17】. However, bronchial wash samples showed a discordant trend, with no significant difference between Liquid-Based Cytology and Conventional smears—diverging from Madhu et al., who reported a clear Liquid-Based Cytology advantage【12】.

5. CONCLUSION

This study highlights a high level of concordance between Liquid-Based Cytology findings and established literature, supporting its superior diagnostic utility over conventional techniques. The discordance observed in the architectural assessment of bronchial samples warrants further exploration, but does not detract from the overall diagnostic gains offered by Liquid-Based Cytology in effusion cytology

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